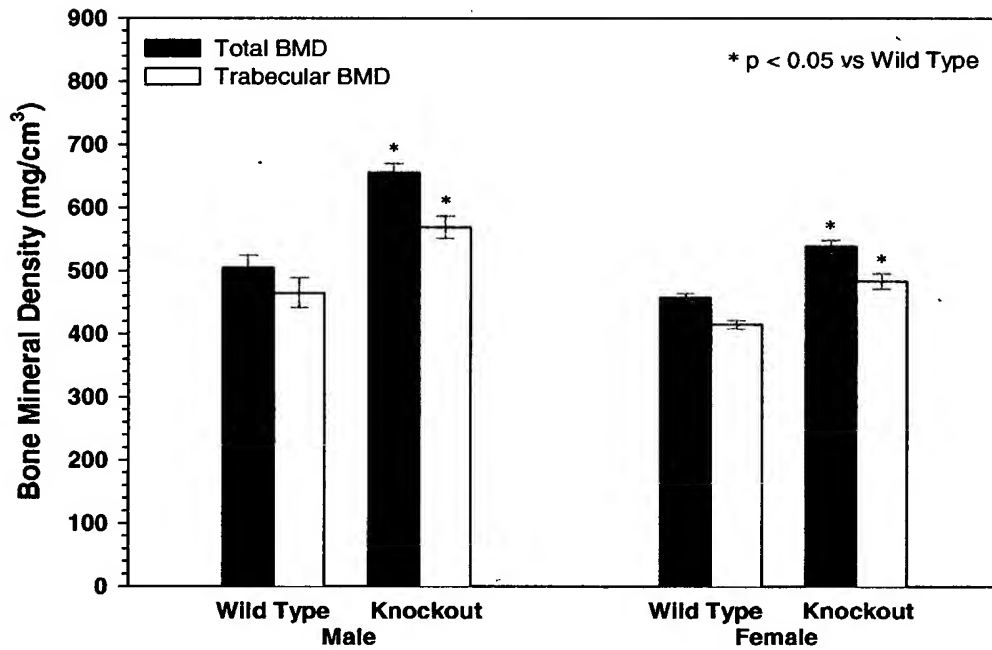
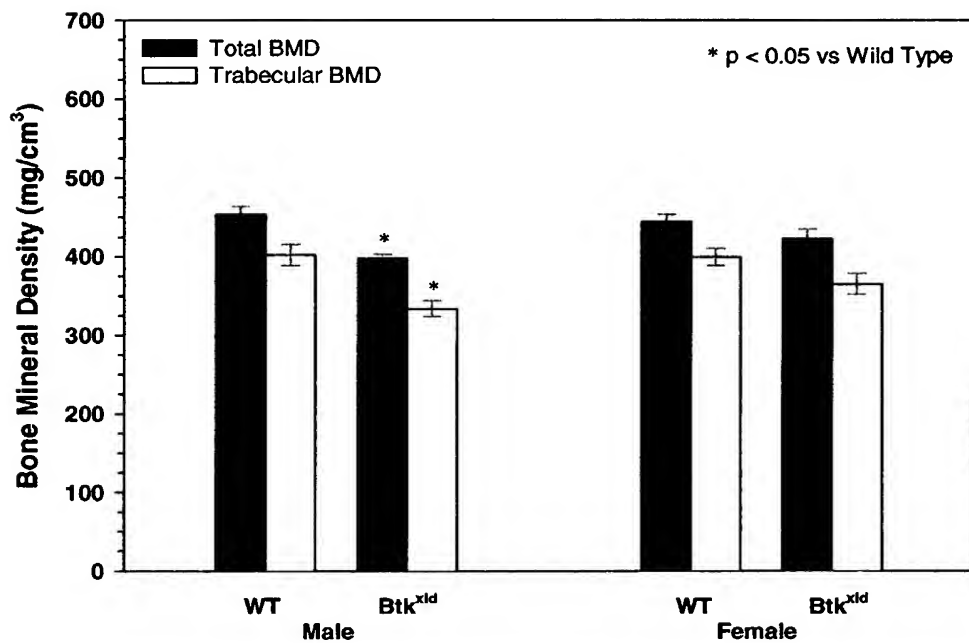


### Bone Mineral Density of the Proximal Tibia in Wild Type and Btk Knockout Mice



**FIG. 1**

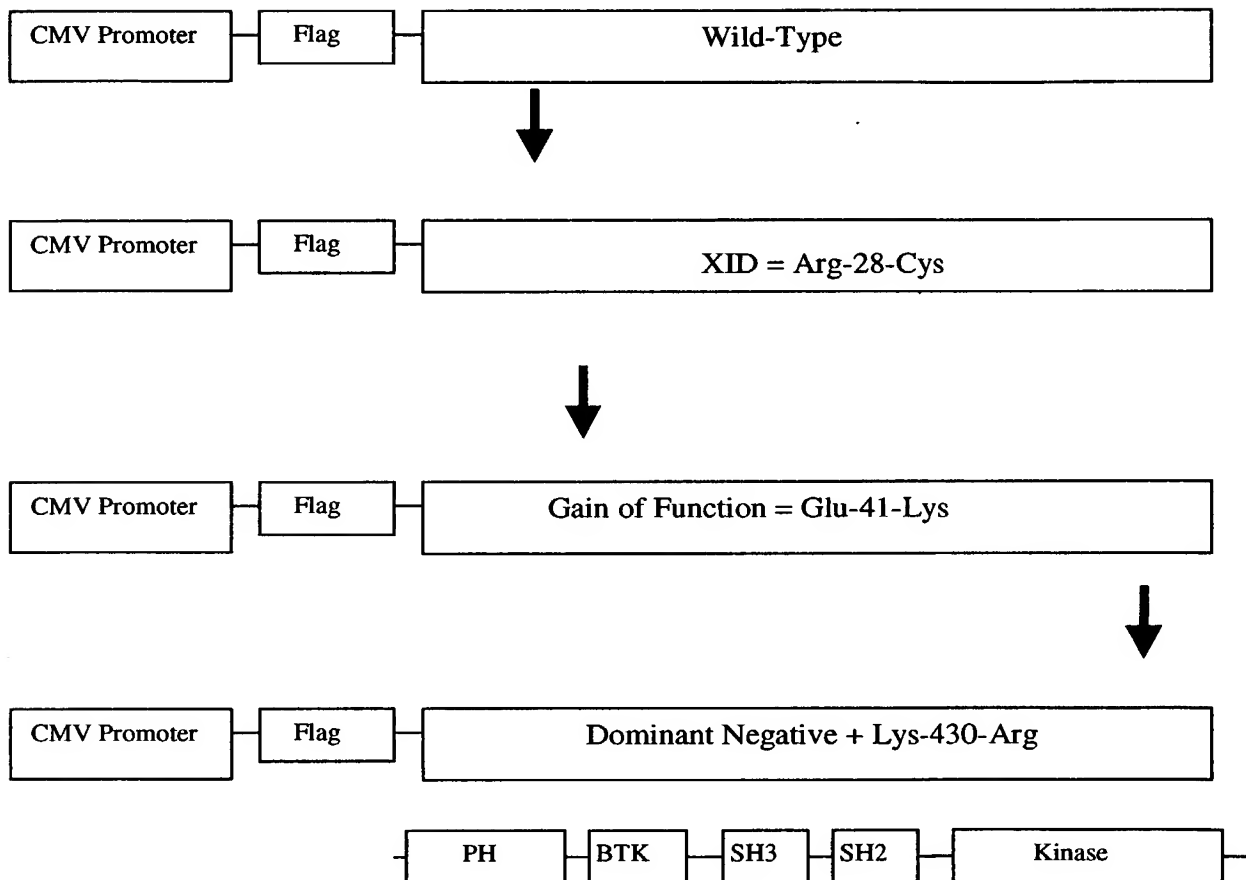
**Bone Mineral Density of the Proximal Tibia  
in Wild Type and Btk<sup>xld</sup> Mice**



**FIG. 2**

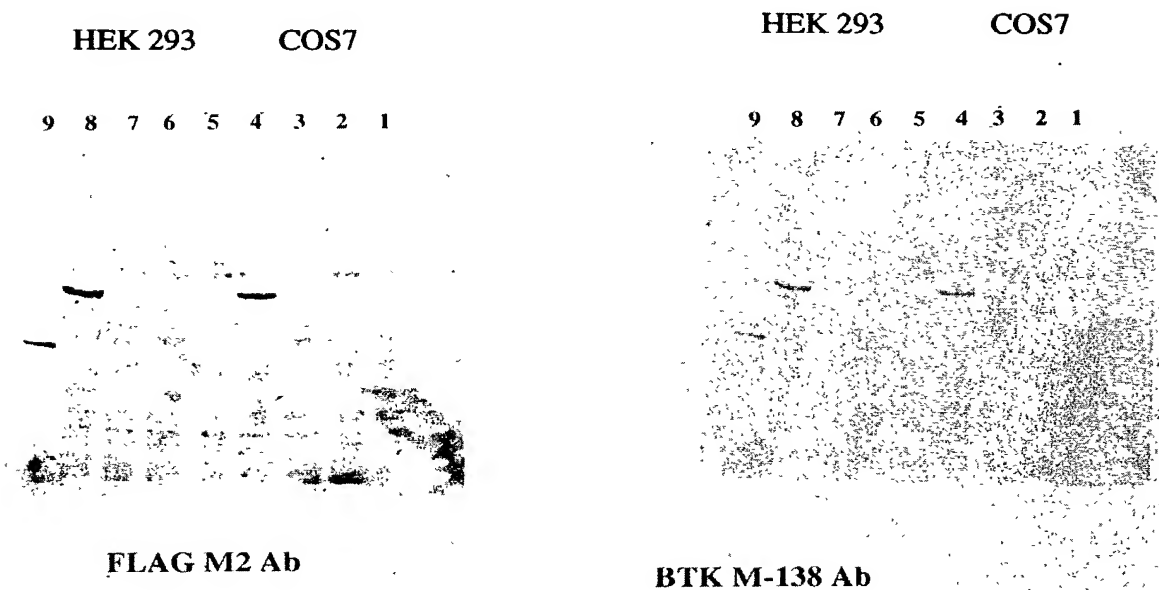


### BTK Constructs



**FIG. 4**

**Mu BTK wt tagged/untagged HEK/COS transluent Western**



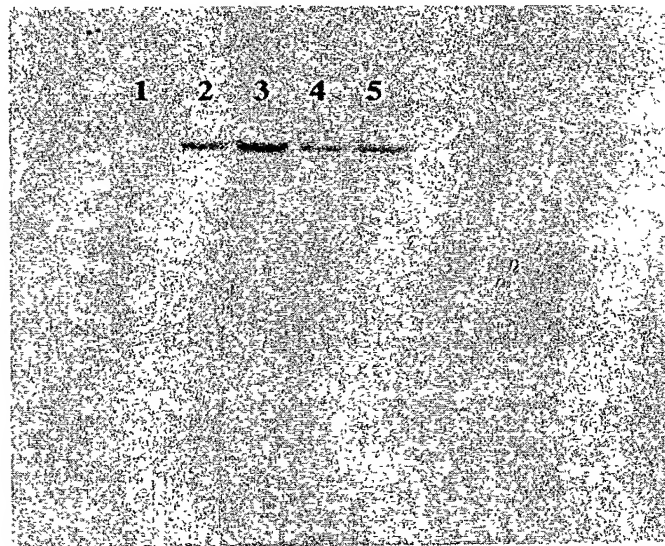
**Legend:**

Lanes 1 and 5: pcDNA  
Lanes 2 and 6: BTK wt/pcDNA  
Lanes 3 and 7: p3XFLAG  
Lanes 4 and 8: BTK wt/p3XFLAG  
Lane 9: BAP FLAG control

**FIG. 5**



**BTK-FLAG mutant RAW 264.7 stable mix whole cell lysates**



$\alpha$ -FLAG Ab

Legend:

- Lane 1: p3xFLAG vector control
- Lane 2: wt BTK in p3xFLAG
- Lane 3: R28C btk in p3XFLAG
- Lane 4: E41K btk in p3XFLAG
- Lane 5: K430R btk in p3XFLAG

**FIG. 6**

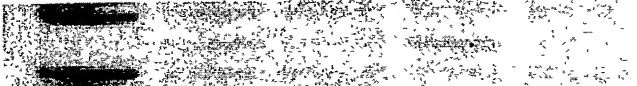


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### Phosphorylation Analysis of BTK Mutants

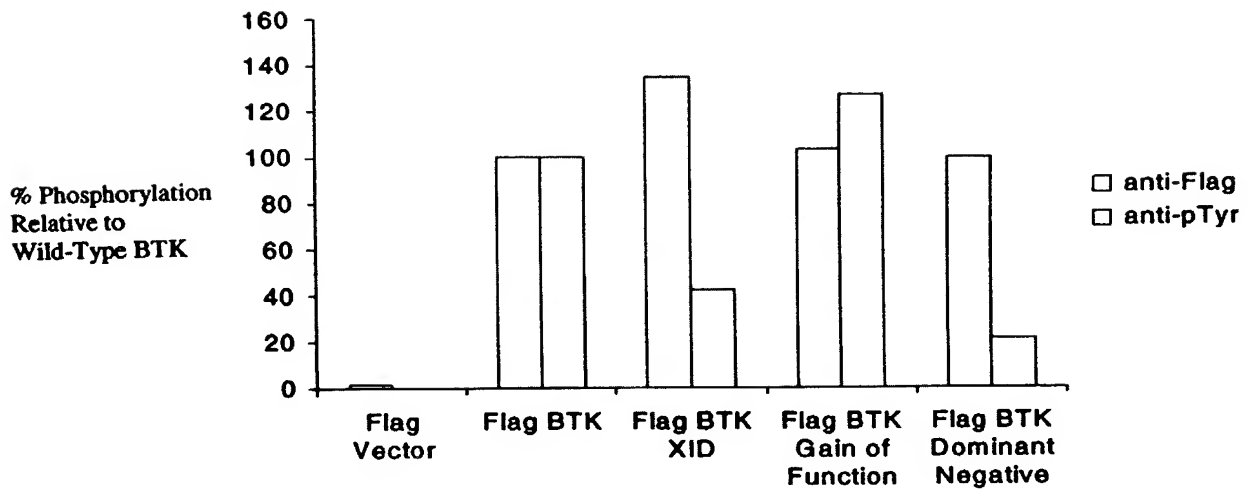
$\alpha$ -Flag  ← Flag-BTK

**FIG. 7a**

$\alpha$ -pTyr  ← Flag-BTK

Flag WT R28C E41K K430R

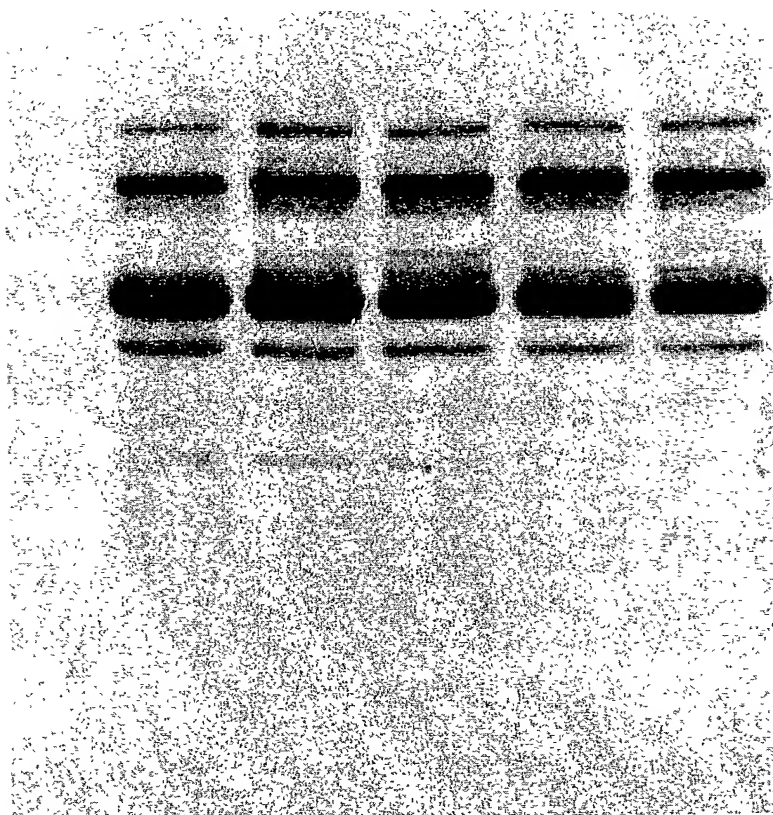
**FIG. 7b**



**FIG. 7c**

### Total Tyrosine Phosphorylation

Vector WT XID G of F DN



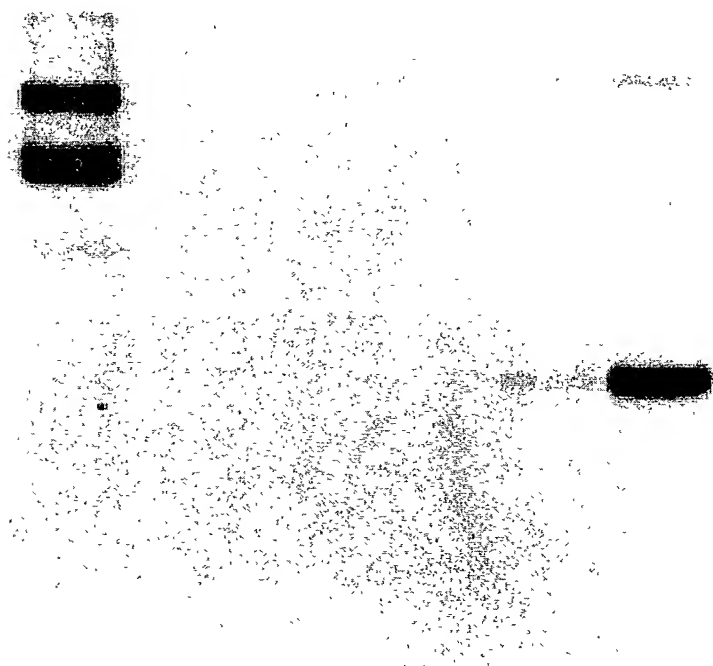
**FIG. 8**





**BTK mutant immunoprecipitation kinase assays**  
*(stable pools, unstimulated)*

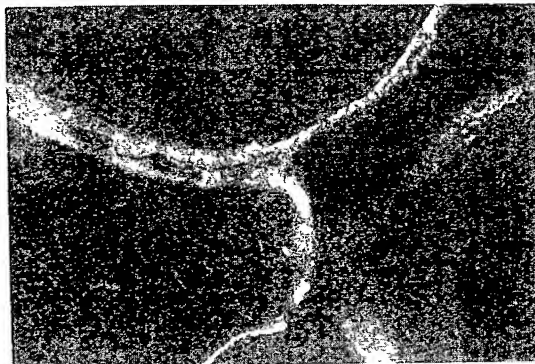
**rBTK    SLP76    Mock IP    Vector    Wild Type    R28C (xid mutant)    E41K    K430R**



**FIG. 9**

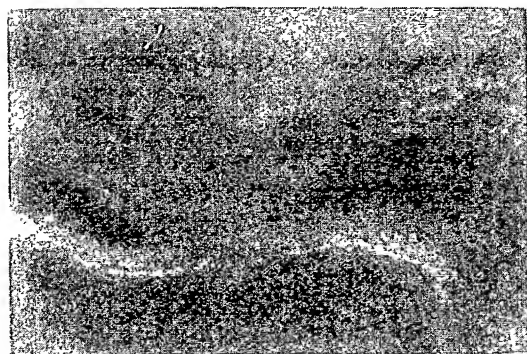
**Actin staining of Btk/RAW cell stable cell lines**

**Btk wt**



**FIG. 10a**

**R28C-xid**



**FIG. 10b**

**E41K-gain of function**



**FIG. 10c**